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=> s acyl-acp desaturase and (mutant? or modified or variant?) and (114 or 117
or 118 or 179 or 181 or 188 or 189)

L1            5 ACYL-ACP DESATURASE AND (MUTANT? OR MODIFIED OR VARIANT?) AND
                (114 OR 117 OR 118 OR 179 OR 181 OR 188 OR 189)

=> dup rem l1

PROCESSING COMPLETED FOR L1
L2            2 DUP REM L1 (3 DUPLICATES REMOVED)

=> d l2 1-2 ibib ab


L2   ANSWER 1 OF 2  CAPLUS  COPYRIGHT 2000 ACS
ACCESSION NUMBER:         1998:31112  CAPLUS
DOCUMENT NUMBER:          128:99301
TITLE:                    Engineered acyl-ACP desaturases with modified
                          chain length and double bond specificity
INVENTOR(S):              Cahoon, Edgar B.; Shanklin, John; Lindgvist, Ylva;
                          Schneider, Gunter
PATENT ASSIGNEE(S):       Associated Universities, Inc., USA
SOURCE:                   U.S., 14 pp.
                          CODEN: USXXAM
DOCUMENT TYPE:            Patent
LANGUAGE:                 English
FAMILY ACC. NUM. COUNT:   2
PATENT INFORMATION:

     PATENT NO.       KIND  DATE           APPLICATION NO.  DATE
```

US 5705391	A	19980106	US 1996-689823	19960814
US 5888790	A	19990330	US 1997-853979	19970509
WO 9806735	A1	19980219	WO 1997-US13690	19970801
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, UZ, VN, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9738266	A1	19980306	AU 1997-38266	19970801
AU 718200	B2	20000406		
EP 934332	A1	19990811	EP 1997-935294	19970801
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI				
BR 9711078	A	20000111	BR 1997-11078	19970801
NO 9900669	A	19990412	NO 1999-669	19990212
US 6100091	A	20000808	US 1999-276295	19990325
PRIORITY APPLN. INFO.:			US 1996-689823	19960814
			US 1997-853979	19970509
			WO 1997-US13690	19970801

AB Disclosed is a methods for modifying the chain length and double bond positional specificities of a sol. plant fatty acid desaturase. More specifically, the method involves modifying amino acid contact residues in the substrate binding channel of the sol. fatty acid desaturase which contact the fatty acid. Amino acid contact residues which lie within the substrate binding channel are identified by alignment with the primary amino acid sequence of the Ricinus communis .DELTA.9 desaturase for max. sequence conservation. A 3-dimensional model for the acyl-[acyl carrier protein] desaturase is then constructed based on the sequence conservation with the R. communis .DELTA.9 desaturase. A **mutant acyl -ACP desaturase** having **modified** chain length and double bond positional specificity is then generated by replacing one or more of the amino acid contact residues with another amino acid residue. Residues at conserved positions **114**, 115, **117**, **118**, **179**, **181**, **188**, and **189** are most relevant in detg. specificity of the desaturase.

L2 ANSWER 2 OF 2 MEDLINE DUPLICATE 1

ACCESSION NUMBER:	1998289105 MEDLINE
DOCUMENT NUMBER:	98289105
TITLE:	A determinant of substrate specificity predicted from the acyl-acyl carrier protein desaturase of developing cat's claw seed.
AUTHOR:	Cahoon E B; Shah S; Shanklin J; Browse J
CORPORATE SOURCE:	Biology Department, Brookhaven National Laboratory, Upton, New York 11976, USA.
SOURCE:	PLANT PHYSIOLOGY, (1998 Jun) 117 (2) 593-8. Journal code: P98. ISSN: 0032-0889.
PUB. COUNTRY:	United States
	Journal; Article; (JOURNAL ARTICLE)
LANGUAGE:	English
FILE SEGMENT:	Priority Journals
ENTRY MONTH:	199810
ENTRY WEEK:	19981001

AB Cat's claw (Doxantha unguis-cati L.) vine accumulates nearly 80% palmitoleic acid (16:1Delta9) plus cis-vaccenic acid (18:1Delta11) in its seed oil. To characterize the biosynthetic origin of these unusual fatty acids, cDNAs for acyl-acyl carrier protein (acyl-ACP) desaturases were isolated from developing cat's claw seeds. The predominant **acyl-ACP desaturase** cDNA identified encoded a polypeptide that is closely related to the stearoyl (Delta9-18:0)-ACP desaturase from castor (Ricinis communis L.) and other species. Upon expression in Escherichia coli, the cat's claw polypeptide functioned as a Delta9 **acyl-ACP desaturase** but displayed a distinct substrate specificity for palmitate (16:0)-ACP rather than stearate (18:0)-ACP. Comparison of the predicted amino acid sequence of the cat's claw enzyme with that of the castor Delta9-18:0-ACP desaturase suggested that a single amino acid substitution (L118W) might account in large part for the differences in substrate specificity between the two desaturases. Consistent with this prediction, conversion of leucine-**118** to tryptophan in the mature castor Delta9-18:0-ACP desaturase resulted in an 80-fold increase in the relative specificity of this enzyme for 16:0-ACP. The alteration in substrate specificity observed in the L118W **mutant** is in agreement with a crystallographic model of the proposed substrate-binding pocket of the castor Delta9-18:0-ACP desaturase.

=> s acyl-acp desaturase and (mutant? or modified or variant?) and (gene or dna or nucleic acid)

4 FILES SEARCHED...
L3 3 ACYL-ACP DESATURASE AND (MUTANT? OR MODIFIED OR VARIANT?) AND (GENE OR DNA OR NUCLEIC ACID)

=> dup rem l3

PROCESSING COMPLETED FOR L3
L4 2 DUP REM L3 (1 DUPLICATE REMOVED)

=> d l4 ibib ab

L4 ANSWER 1 OF 2 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 1998289105 MEDLINE
DOCUMENT NUMBER: 98289105
TITLE: A determinant of substrate specificity predicted from the acyl-acyl carrier protein desaturase of developing cat's claw seed.
AUTHOR: Cahoon E B; Shah S; Shanklin J; Browse J
CORPORATE SOURCE: Biology Department, Brookhaven National Laboratory, Upton, New York 11976, USA.
SOURCE: PLANT PHYSIOLOGY, (1998 Jun) 117 (2) 593-8.
Journal code: P98. ISSN: 0032-0889.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199810
ENTRY WEEK: 19981001
AB Cat's claw (Doxantha unguis-cati L.) vine accumulates nearly 80% palmitoleic acid (16:1Delta9) plus cis-vaccenic acid (18:1Delta11) in its seed oil. To characterize the biosynthetic origin of these unusual fatty

acids, cDNAs for acyl-acyl carrier protein (acyl-ACP) desaturases were isolated from developing cat's claw seeds. The predominant **acyl-ACP desaturase** cDNA identified encoded a polypeptide that is closely related to the stearoyl (Delta9-18:0)-ACP desaturase from castor (Ricinis communis L.) and other species. Upon expression in Escherichia coli, the cat's claw polypeptide functioned as a Delta9 **acyl-ACP desaturase** but displayed a distinct substrate specificity for palmitate (16:0)-ACP rather than stearate (18:0)-ACP. Comparison of the predicted amino acid sequence of the cat's claw enzyme with that of the castor Delta9-18:0-ACP desaturase suggested that a single amino acid substitution (L118W) might account in large part for the differences in substrate specificity between the two desaturases. Consistent with this prediction, conversion of leucine-118 to tryptophan in the mature castor Delta9-18:0-ACP desaturase resulted in an 80-fold increase in the relative specificity of this enzyme for 16:0-ACP. The alteration in substrate specificity observed in the L118W **mutant** is in agreement with a crystallographic model of the proposed substrate-binding pocket of the castor Delta9-18:0-ACP desaturase.

=> d l4 ibib ab 2

L4 ANSWER 2 OF 2 MEDLINE
ACCESSION NUMBER: 97289686 MEDLINE
DOCUMENT NUMBER: 97289686
TITLE: Redesign of soluble fatty acid desaturases from plants for altered substrate specificity and double bond position.
AUTHOR: Cahoon E B; Lindqvist Y; Schneider G; Shanklin J
CORPORATE SOURCE: Biology Department, Brookhaven National Laboratory, Upton, NY 11973, USA.
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1997 May 13) 94 (10) 4872-7. Journal code: PV3. ISSN: 0027-8424.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199708
ENTRY WEEK: 19970801

AB Acyl-acyl carrier protein (ACP) desaturases introduce double bonds at specific positions in fatty acids of defined chain lengths and are one of the major determinants of the monounsaturated fatty acid composition of vegetable oils. Mutagenesis studies were conducted to determine the structural basis for the substrate and double bond positional specificities displayed by acyl-ACP desaturases. By replacement of specific amino acid residues in a Delta6-palmitoyl (16:0)-ACP desaturase with their equivalents from a Delta9-stearoyl (18:0)-ACP desaturase, **mutant** enzymes were identified that have altered fatty acid chain-length specificities or that can insert double bonds into either the Delta6 or Delta9 positions of 16:0- and 18:0-ACP. Most notably, by replacement of five amino acids (A181T/A200F/S205N/L206T/G207A), the Delta6-16:0-ACP desaturase was converted into an enzyme that functions principally as a Delta9-18:0-ACP desaturase. Many of the determinants of fatty acid chain-length specificity in these **mutants** are found in residues that line the substrate binding channel as revealed by x-ray crystallography of the Delta9-18:0-ACP desaturase. The crystallographic model of the active site is also consistent with the diverged activities

associated with naturally occurring **variant** acyl-ACP desaturases. In addition, on the basis of the active-site model, a Delta9-18:0-ACP desaturase was converted into an enzyme with substrate preference for 16:0-ACP by replacement of two residues (L118F/P179I). These results demonstrate the ability to rationally modify **acyl-ACP desaturase** activities through site-directed mutagenesis and represent a first step toward the design of acyl-ACP desaturases for the production of novel monounsaturated fatty acids in transgenic oilseed crops.

=> d his

(FILE 'HOME' ENTERED AT 16:01:42 ON 16 NOV 2000)

FILE 'MEDLINE, CAPLUS, EMBASE, BIOSIS, SCISEARCH' ENTERED AT 16:02:21 ON 16 NOV 2000

L1 5 S ACYL-ACP DESATURASE AND (MUTANT? OR MODIFIED OR VARIANT?)
AND
L2 2 DUP REM L1 (3 DUPLICATES REMOVED)
L3 3 S ACYL-ACP DESATURASE AND (MUTANT? OR MODIFIED OR VARIANT?)
AND
L4 2 DUP REM L3 (1 DUPLICATE REMOVED)

=> log

ALL L# QUERIES AND ANSWER SETS ARE DELETED AT LOGOFF
LOGOFF? (Y)/N/HOLD:y

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